

Claims

1. Reverse transcriptase (RT) assay kit comprising one or several package(s) containing solid phase bound polyriboadenylic acid (prA) and/or polydeoxyadenylic acid (pdA) template(s) obtainable by contacting a polystyrene-based solid phase with a coupling solution comprising 1-methylimidazole, and prA and/or pdA, followed by incubation, washing with a wash buffer, drying and packaging,

RT-type adapted separately packaged assay components selected from the group consisting of a mixture of or separately a buffer, $\text{pH} \approx 7-8$, divalent metal ion, chelator, polyamine, RNase inhibitor, reducing agent, salt, stabilizing agent, and detergent, and a mixture of or separately lyophilized deoxynucleotide triphosphate, primer, protective agent and a concentrated washing buffer, and optionally lyophilized reference enzyme(s), and optionally components of a detection system comprising lyophilized alkaline phosphatase conjugated anti-BrdU monoclonal antibody, alkaline phosphatase substrate buffer and alkaline phosphatase substrate, and written instructions for use of the assay kit.

2. RT assay kit according to claim 1, wherein the solid phase is a microtiter plate and an aliquot of the coupling solution, which comprises 100 mM 1-methylimidazole, $\text{pH} \approx 5-7$, and 0.5 - 2 mg/ml prA and/or pdA, is added to each well, followed by the incubation at a temperature of 10 - 60 °C for 0.5 - 10 h, and washing each well for the removal of the 1-methylimidazole with the wash buffer, which comprises Bis-Tris propane, $\text{pH} \approx 5-7$, and drying and packaging the plates.

3. RT assay kit according to claim 2, wherein 100 μl of the coupling solution, which comprises 100 mM 1-methylimidazole, $\text{pH} \approx 6.25$, and 1mg/ml prA and/or pdA, is added to each well, followed by the incubation at room temperature for ≈ 2 h, washing of each well with 2x300 μl of the wash buffer, which comprises 10mM Bis-Tris propane, $\text{pH} \approx 6.25$, drying the plates at 37 °C for ≈ 25 minutes and putting the plates in foil bags and vacuum sealing the bags.

4. RT assay kit according to any one of claims 1 - 3, wherein the assay components are selected from the group consisting of the buffers Tris and Hepes, $\text{pH} \approx 7-8$, the divalent metal ions Mg^{2+} and Mn^{2+} , the chelators ethylenediaminetetraacetic acid (EDTA) and ethylene glycol-bis(β -aminoethyl ether) N,N,N',N'-tetraacetic acid (EGTA) and, the

polyamines spermine and spermidine, the RNase inhibitors heparin sulfate and dextran sulfate, the reducing agents dithiothreitol (DTT), dithioerythritol (DTE), and glutathione, the salts NaCl and KCl, the stabilizing agents newborn calf serum (NCS) and bovine serum albumin (BSA), the detergents Tween 20 and Triton X-100, the deoxynucleotide triphosphate BrdUTP, the primer oligo dT, and the protective agent agents ATP, GTP and CTP.

5 5. Use of an assay kit according to any one of claims 1 - 4 for the qualitative and quantitative analysis of RT activity in a biological sample.

6. Use according to claim 5, wherein the biological sample is selected from biological fluids and cell extracts. *a*

10 7. Use according to claim 6, wherein the biological fluid is selected from plasma, serum, spinal fluid, synovial fluid and pleural fluid.

8. Use according to any one of claims 5 - 7 followed by evaluation of the status of a RT activity related disorder or disease based on the result of the analysis of the RT activity.

15 9. Method of qualitative and quantitative analysis of RT activity in a biological sample comprising the steps of using and following the written instructions for the RT assay kit according to any one of claims 1 - 4 for the determination of the RT activity in the biological sample. *a2*

20 10. Method according to claim 9, wherein the biological sample is selected from biological fluids and cell extracts.

11. Method according to claim 10, wherein the biological fluid is selected from plasma, serum, spinal fluid, synovial fluid and pleural fluid.

a3 25 12. Method according to any one of claims 9 - 11 followed by evaluation of the status of a RT activity related disorder or disease based on the result of the analysis of the RT activity.

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